SHORT COMMUNICATION

Chemical constituents of the essential oil and organic acids from longkong (*Aglaia dookkoo* Griff.) fruits

Vanida Chairgulprasert[1], Boonsong Krisornpornsan[2] and Abdulhakim Hamad[3]

Abstract

Chairgulprasert, V., Krisornpornsan, B. and Hamad, A.
Chemical constituents of the essential oil and organic acids from longkong (*Aglaia dookkoo* Griff.) fruits
Songklanakarin J. Sci. Technol., 2006, 28(2) : 321-326

The pulp of longkong fruits (*Aglaia dookkoo* Griff.), collected from Narathiwat province, was dried and extracted by steam distillation to obtain the essential oil in 0.48% yield. The GC-MS data showed oleic acid (14.80%), α-copaene (11.15%), germacrene-D (9.16%), δ-cadinene (6.74%), τ -muurolol (6.34%), (+) spathulenol (5.72%) and palmitic acid (5.49%) as the major constituents. Organic acids were also extracted from dried pulp with methanol using a Soxhlet apparatus to give the crude extract in 36.26% yield. Four organic acids: glycolic, maleic, malic and citric acids were determined by HPLC. Maleic acid (1.23%) was the major acid and the others were citric (0.22%), malic (0.15%) and glycolic acids (0.14%).

Key words : longkong, *Aglaia dookkoo*, essential oil, organic acid

[1]Ph.D.(Chemistry), [3]B.Sc. student in Industrial Chemistry, Department of Science, [2]M.Sc.(Agriculture), Asst. Prof., Department of Technology and Industry, Faculty of Science and Technology, Prince of Songkla University, Muang, Pattani 94000, Thailand.
Corresponding e-mail : cvanida@bunga.pn.psu.ac.th
Received, 12 May 2005 Accepted, 18 July 2005

บทคัดย่อ

วนิดา เจียรกุลประเสริฐ[1] บุญส่ง ไกรสรพรรสร[2] และ อับดุลฮากีม หามะ[1]

องค์ประกอบทางเคมีของน้ำมันหอมระเหยและกรดอินทรีย์จากผลลองกอง (*Aglaia dookkoo* Griff.)

ว. สงขลานครินทร์ วทท. 2549 28(2) : 321-326

การศึกษาองค์ประกอบทางเคมีของน้ำมันหอมระเหย และกรดอินทรีย์ของผลลองกอง (*Aglaia dookkoo* Griff.) จากจังหวัดนราธิวาส พบว่าเมื่อนำเนื้อลองกองแห้งมากลั่นด้วยไอน้ำ ได้น้ำมันหอมระเหยสีเหลืองอ่อนปริมาณ 0.48% และจากการวิเคราะห์ด้วยเครื่อง GC-MS พบสารประกอบหลัก คือ oleic acid (14.80%), α-copaene (11.15%), germacrene-D (9.16%), δ-cadinene (6.74%), τ-muurolol (6.34%), (+) spathulenol (5.72%) และ palmitic acid (5.49%) ในการสกัดกรดอินทรีย์จากเนื้อลองกองแห้งด้วยเมทานอลโดยใช้ชุดสกัดซอกฮ์เลตได้สารสกัดหยาบ 36.26% และจากการวิเคราะห์ด้วยเครื่องโครมาโทกราฟีของเหลวสมรรถนะสูง พบกรดอินทรีย์ 4 ชนิด โดยมี maleic acid ในปริมาณสูงสุด 1.23% ตามด้วย citric acid, malic acid และ glycolic acid 0.22, 0.15 และ 0.14% ตามลำดับ

[1]ภาควิชาวิทยาศาสตร์ [2]ภาควิชาเทคโนโลยีและการอุตสาหกรรม มหาวิทยาลัยสงขลานครินทร์ อำเภอเมือง จังหวัดปัตตานี 94000

Longkong (*Aglaia dookkoo* Griff.), a well-known fruit of Thailand, belonging to the Meliaceae family, has its origin in the South of Thailand, Indonesia, the Philippines and the Malau Islands. It is an economically important plant of Peninsula Thailand and widely distributed throughout the South and East of the Peninsula. Longkong is also cultivated in Australia, Sri Lanka, Vietnam, Burma, India and Puerto Rico (Paull, 2004). Longkong pulp is juicy with an aromatic smell and a sweet but slightly sour taste. The fruits contain a variety of nutrients, including protein and carbohydrates and combine low fat with a high percentage of vitamins and minerals (Sabah, 2004). Moreover, the fruit peel and seeds have various uses in traditional medicine (Verheij and Coronel, 1992). One of the most important quality standards for fruit products is their chemical constituents. Organic acids have a significant influence on the characteristic fruit flavor, stability and colour (Ryan and Dupont, 1973; Romero and Munoz, 1993). In general, fruits contain a variety of organic acids. For example, grape contains citric, malic and tartaric acids (Soyer *et al*., 2003), while strawberry contains malic and citric acids, apple contains malic acid (Fuso, 1998) and green mango from Bengal contains tartaric, citric and maleic acids (Bangalinet, 2001-2003). Essential oils affect the organoleptic profiles of fruits. They are found in diverse aromatic fruits such as *Annona squamosa* (Andrade *et al*., 2001), *Protium icicariba* (Siani *et al*., 2004) and *Citrus cultivars* (Merle *et al*., 2004).

Apart from analyses of nutritional components as described above, no study has been made of other chemical constituents of the longkong fruit. Therefore, this is the first report on the investigation of the chemical constituents of the essential oil and organic acids from longkong.

Materials and Methods

Plants, Materials and Chemicals

Longkong fruits were harvested from Seepoa village, Narathiwat province. They were picked at the ripe mature stage, washed and separated from their bunches. After the skin and seeds were separated, the pulp was dried at 50°C.

A reference standard of glycolic, malic and maleic acids was purchased from Fluka (Buchs, Switzerland). Lactic acid was obtained from Vidhyasom (Bangkok, Thailand) and citric acid from Ajax Finechem (NSW, Australia). Standard organic acids were used without purification. All reagents and solvents were analytical grade and used as received.

Stock solutions of glycolic, malic, lactic,

maleic and citric acids were prepared in deionized water at a concentration of 1000 ppm. Standard solutions were further diluted with deionized water to the concentration range of 25-200 ppm and filtered through a 0.45 μm membrane.

Essential oil extraction

The dried longkong pulp (100 g) was ground in a blender. The resulting material was subjected to steam distillation for 4 h using a Dean-Stark apparatus. The obtained essential oil (0.48 g) was dried over anhydrous sodium sulfate and stored at -20ºC prior to analysis. Constituents of the essential oil were analyzed by GC-MS.

Organic acid extraction

The dried pulp (100 g) was ground in a blender and extracted with methanol in a Soxhlet apparatus for 7 h. The solvent was evaporated under reduced pressure to yield the crude extract (36.26 g). The extract (0.10 g) was added to a 5 mL solution of disodium hydrogenphosphate and the mixture was sonicated for 15 minutes and adjusted to a volume of 25 mL with deionized water. The supernatant was filtered through a 0.45 μm membrane and injected into the HPLC for analysis of glycolic, malic, maleic, lactic and citric acids.

GC-MS analysis

GC-MS analysis was performed on a Hewlet-Packard 5890 gas chromatograph, coupled to a HP 5972 mass selective detector. The GC was fitted with an Rtx-5MS column (30 m x 0.25 mm I.D., film thickness 0.25 μm). The inlet temperature was set at 280ºC and the oven temperature was programmed from 80ºC to 300ºC at 7ºC/min rate and held at this temperature for 5 min. Helium was used as carrier gas. The mass spectrometer was run in electron ionization mode, scanning at 45-550 amu. A solvent delay time was 3 min with a transfer line temperature of 300ºC and the sample injection volume was 1 μL. The relative proportion of each individual component of the oil was expressed as a percentage relative to the total peak area.

HPLC analysis

The HPLC system comprised a 717 Plus autosampler, a Water 510 pump (Millipore) and a Water 996 photodiode array. The separation was carried out on a Hypersil ODS reverse phase column (5 μm, 40 x 250 mm) (Hewlett Packard). Injection volume was 10 μL with an isocratic flow rate 0.5 mL/min. and UV detector was set at 210 nm. Mobile phase was 0.05 M disodium hydrogen-phosphate, adjusted with hydrochloric acid to pH 2.5. The mobile phase was filtered through a 0.45 μm membrane and degassed before use.

Glycolic, malic, maleic, lactic and citric acids were identified by comparing their retention times with those of the corresponding standards. The quantitative analyses were carried out using linear calibration graphs from standard solutions of authentic compounds in the concentration range of 25-200 ppm. Each dilution was carried out in triplicate and the mean value was used.

Results and Discussion

Determination of the essential oil

The essential oil from dried longkong pulp was obtained as a pale yellow oil with an aromatic smell in 0.48 % yield. According to GC-MS analysis, 20 compounds were identified. The relative percentages of these compounds are recorded in Table 1. The main constituents were oleic acid (14.80%), α- copaene (11.15%), germacrene-D (9.16%), δ-cadinene (6.74%), τ-muurolol (6.34%), (+) spathulenol (5.72%) and palmitic acid (5.49%). Most of the essential oil components were sesquiterpenes (69.81%) and only one monoterpene, linalool (0.80%) was present. In addition, two fatty acids, oleic acid and palmitic acid, were detected.

Determination of organic acids

In this study, 5 organic acids including glycolic, malic, maleic, lactic and citric acids were qualitatively and quantitatively analysed by HPLC. A chromatogram of a mixture of standard acids is shown in Figure 1A. The retention times of glycolic, malic, lactic, maleic and citric acids were 5.5, 6.5,

Table 1. Chemical constituents of the essential oil from longkong pulp.

Peak No.	Compound	% of Total
1	L-Linalool	0.80
2	α-Copaene	11.15
3	2,4-diisopropenyl-1-methyl-1-vinylcyclohexane	1.25
4	β-Caryophyllene	1.46
5	γ-Muurolene	4.56
6	Germacrene-D	9.16
7	α-Muurolene	3.69
8	γ-Cadinene	2.42
9	δ-Cadinene	6.74
10	α-Calacorene	2.02
11	(+)-Spathulenol	5.72
12	Aromadendrene	2.84
13	Ledane	4.09
14	Fonenol	2.05
15	α-Longipinene	1.56
16	Torreyol	4.58
17	τ-Muurolol	6.34
18	(2,3,4-Trimethylphenyl)-2-propanone	2.12
19	Palmitic acid	5.49
20	*cis*-Oleic acid	14.80

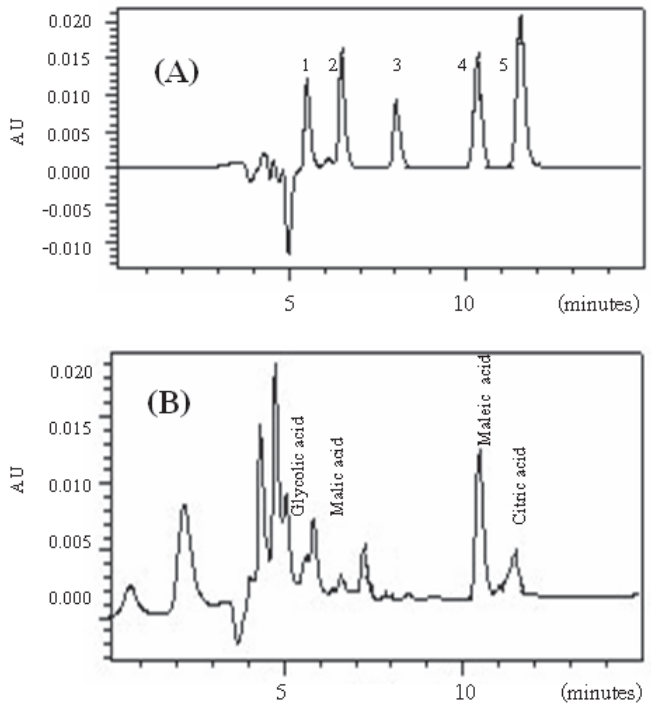

Figure 1. HPLC Chromatogram of (A) standard mixture of organic acids: (1) glycolic acid (2) malic acid (3) lactic acid (4) maleic acid (5) citric acid and (B) organic acids in the sample.

8.0, 10.4 and 11.5 minutes, respectively. After injection of the crude sample, the chromatogram was recorded as shown in Figure 1B.

From a comparison between the retention time of components in the standard and sample chromatograms, it was found that 4 signals from the sample corresponded to glycolic, malic, maleic and citric acids. No lactic acid peak was detected. These results were confirmed by spiking with a small amount of each acid from a standard stock solution and testing individually. In quantitative analyses, the calibration curves were compared over the concentration range 25-200 ppm. Each point of the calibration graph corresponded to the mean value from three independent peak measurements. The calibration graphs of each standard were linear with correlation coefficient greater than 0.99 (Figure 2a-d). Based upon the graphs, maleic acid was identified as the major acid, accounting for 1.23 % w/w of dried pulp. Citric, malic and glycolic acids were present at the levels of 0.22, 0.15 and 0.14 % w/w of dried pulp (Table 2). Maleic acid can promote fruit aroma and develop taste in food and beverages. It has become a new acidulant for the food and beverage industry (Changmao Biochem, 1999-2003).

Conclusions

In summary, the significant characteristic fragrance of longkong fruit, an aroma impression of various essential oil constituents, was identified. In addition, a group of organic acids was identified of which maleic acid is the major acid responsible for a slightly sour flavour. These results provide valuable information for quality control and development of longkong products. Future work in this area will greatly benefit our understanding of the natural fluctuations in the essential oil and organic acid components in longkong fruit depending on harvesting time.

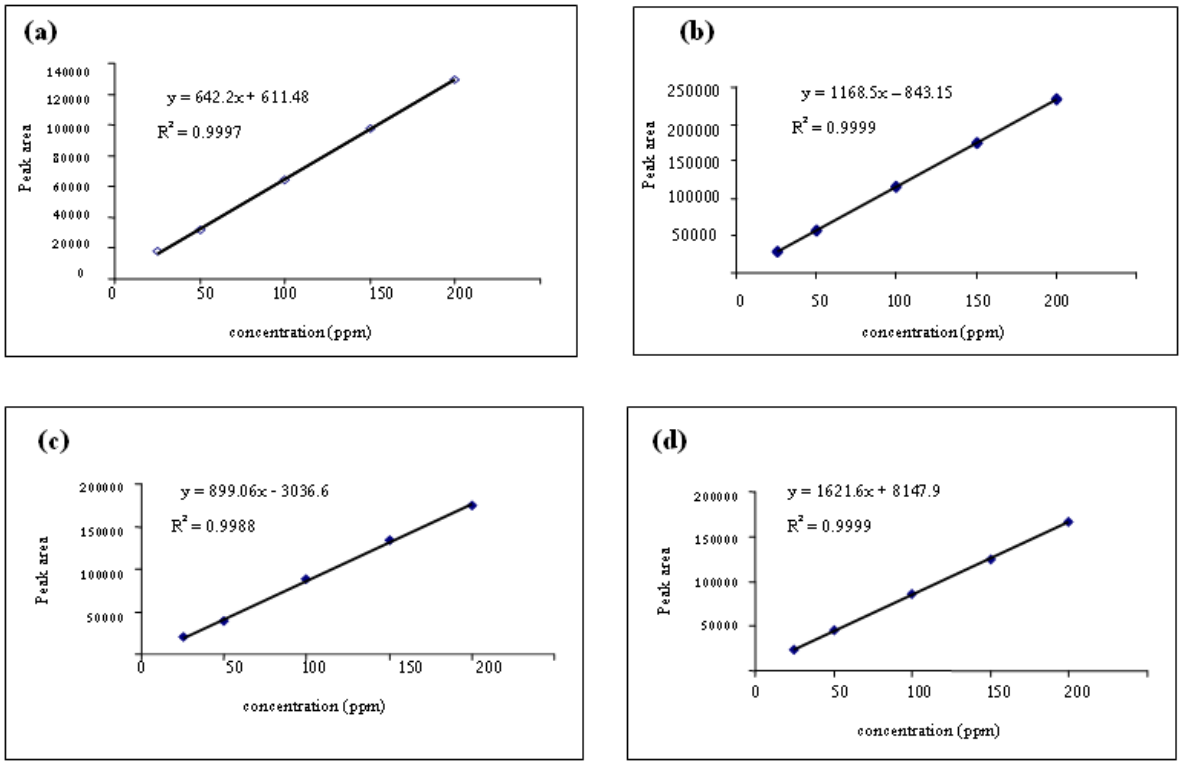

Figure 2. The standard graphs of (a) glycolic acid, (b) maleic acid, (c) malic acid and (d) citric acid.

Table 2. Peak areas and quantities of organic acids in longkong pulp.

Organic acids	Peak area	Quantities of acids (% w/w of dried pulp)
Maleic acid	157938.67	1.23
Citric acid	47915.00	0.22
Malic acid	11878.33	0.15
Glycolic acid	10335.67	0.14

Acknowledgements

We would like to thank Department of Science, Faculty of Science and Technology, Prince of Songkla University, Pattani Campus, for support with chemicals and equipment. We are also grateful to Dr. Ian Beadham for his kind proof-reading of this manuscript.

References

Andrade, E.H.A., Zoghbi, M.G.B., Maia, J.G.S., Fabricius, H. and Marx, F. 2001. Chemical characterization of the fruit of *Annona squamosa* L. Occurring in the Amazon, J. Food. Compos. Anal., 14 : 227-232.

Bangalinet. 2001-2003. Plants. http://www.bangalinet.com/bengal_plants.htm. [November 14, 2004].

Changmao Biochem. 1999-2003. Maleic acid. http ://www.Cmbee.Com/en/product/product6.htm. [November 27, 2004].

Fuso. 1998. Fruit acid. http://www.fusokk.co.jp/products/acid/acid01_e.htm. [September 25, 2004].

Merle, H., Moron, M., Blazquez, A. and Boira, H. 2004. Taxonomical contribution of essential oils in mandarins culivars, Biochem. System. Ecol., 32: 491-497.

Paull, R.E. 2004. Longkong.: http://www.ba.ars.usda.gov/hb66/087/longkong.pdf. [November 7, 2004].

Romero, E.G. and Munoz, G.S. 1993. Determination of organic acid in grape, musts, wines and vinegars by high performance liquid chromatography, J. Chromatog. A., 655: 111-117.

Ryan, J.J. and Dupont, J.A. (1993). Identification and analysis of the major acids from fruit juices and wines, J. Agric. Food. Chem., 21: 45-49.

Sabah (Malaysian Agriculture Department), Langsat, Duku, Duku-Langsat and Dokong. http://www.sabah.gov.my/tani/english/crop_langsat.htm. [November 14, 2004].

Siani, A.C., Garrido, I.S., Monterio, S.S., Carvalho, E. S. and Ramos, M.F.S. 2004. *Protium icicariba* as a source of volatile essence, Biochem. System. Ecol., 32: 477-489.

Soyer, Y., Koca, N. and Karadeniz, F. 2003. Organic acid profile of Turkish white grapes and grape juices, J. Food. Compos. Anal., 16: 629-636.

Verheij, E.W.M. and Coronel, R.E. 1992. Edible fruits and nuts, Plants Resources of South-East Asia No.2 PROSEA, Bogor, Indonesia.